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Bioprocessing of Brewers' Spent Grain Enhances Its Antioxidant Activity: Characterization of Phenolic Compounds and Bioactive Peptides

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Brewers' spent grain (BSG) is the major by-product of the brewing industry which remain largely unutilized despite its nutritional quality. In this study, the effects of fermentation on BSG antioxidant potential were analyzed. A biotechnological protocol including the use of xylanase followed by fermentation with *Lactiplantibacillus plantarum* (*Lactobacillus plantarum*) PU1, PRO17, and H46 was used. Bioprocessed BSG exhibited enhanced antioxidant potential, characterized by high radical scavenging activity, long-term inhibition of linoleic acid oxidation and protective effect toward oxidative stress on human keratinocytes NCTC 2544. Immunolabelling and confocal laser microscopy showed that xylanase caused an extensive cell wall arabinoxylan disruption, contributing to the release of bound phenols molecules, thus available to further conversion through lactic acid bacteria metabolism. To clarify the role of fermentation on the antioxidant BSG potential, phenols were selectively extracted and characterized through HPLC-MS techniques. Novel antioxidant peptides were puri ed and identi ed in the most active bioprocessed BSG.

Keywords: brewers' spent grain, bioprocessing, phenolic compounds, bioactive peptides, antioxidant activity

Abbreviations: ABTS, 2,2-azino-di-(3-ethylbenzthiazoline sulfonate); AX, arabinoxylans; BHT, butylated hydroxytoluene; BSA, bovine serum albumin; BSG, brewers' spent grain; CLSM, confocal laser scanning microscopy; DPPH, 2,2-diphenyl-1-picrylhydrazyl; eBSG, enzyme treated brewers' spent grain; eBSG fH46, enzyme treated brewers' spent grain fermented with *L. plantarum* H46; eBSG fPRO17, enzyme treated brewers' spent grain fermented with *L. plantarum* PRO17; eBSG fPU1, enzyme treated brewers' spent grain fermented with *L. plantarum* PU1; EF30, bioprocessing protocol including simultaneous enzymatic treatment and fermentation at 30°C for 24 h; E50C F30, Bioprocessing protocol including sequential enzymatic treatment at 50°C for 5 h and fermentation at 30°C for 24 h; FBS, fetal bovine serum; HPLC-FLD, high-performance liquid chromatography with fluorescence detection; LAB, lactic acid bacteria; ME, methanolic extracts; MRS, De Man,

experiments. VV and ED-C oversaw MV in the phenolic profile characterization. RC and CR conceived and designed the experimental plan. CR oversaw the writing process. All authors read and approved the final manuscript.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fmicb.2020.01831/full#supplementary-material

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Conflict of Interest: DP and FR are employed by Giuliani S.p.A.

The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.